

o WE CLAIM:

1. A method of delivering antigen to dendritic cells comprising:

5                   contacting dendritic cells with apoptotic cells expressing an antigen wherein said contact is for a time sufficient to allow said antigen to be internalized by the dendritic cells, and wherein said apoptotic cells have been induced in vitro to become apoptotic.

10           2. The method according to claim 1 wherein the dendritic cells are human.

15           3. The method according to claim 1 wherein the apoptotic cells are selected from the group consisting of cells of a cell line, cells which have been transformed to express a foreign antigen, tumor cell line, xenogeneic cells, or tumor cells.

20           4. The method according to claim 3 wherein the apoptotic cells are selected from the group consisting of

                  monocytes, 293 cells, L cells, Hela cells, B cells and EL4 cells.

25           5. The method according to claim 1 further comprising the step of inducing apoptosis of cells expressing said antigen to produce the apoptotic cells.

30           6. The method according to claim 5 wherein apoptosis is induced by infection with influenza virus.

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7. The method according to claim 5 wherein apoptosis is induced by irradiation with ultraviolet light, gamma radiation, steroids, serum deprivation, cytokines, or drugs.
8. The method according to claim 5 wherein apoptosis is induced by depriving antigen donor cells of nutrients in the cell culture medium.
9. The method according to claim 1 wherein dendritic cells are exposed to a preparation of apoptotic cell fragments, blebs, or bodies containing antigen.
10. The method according to claim 1 wherein said antigen is selected from a group consisting of tumor antigens, viral antigens, pathogens, microbial antigens, self antigens, and autoimmune antigens.
11. The method according to claim 10 wherein the antigen is selected from the group consisting of influenza virus, malaria, HIV, EBV, human papilloma virus, CMV, renal cell carcinoma antigens, melanoma antigens, breast cancer antigens, cancer antigens and myeloma antigens.
12. The method according to claim 10 wherein the antigen is a tumor antigen.
13. The method according to claim 1 wherein said dendritic cells are immature and phagocytic.
14. The method according to claim 1 wherein the cells to be induced to undergo apoptosis are first transformed with DNA encoding said antigen.

15. The method according to claim 1 wherein the ratio of apoptotic cells to dendritic cells is about 1-10 apoptotic cells to about 100 dendritic cells.

16. The method according to claim 1 wherein the dendritic cells are contacted with the apoptotic cells in vivo.

17. The method according to claim 1 wherein the dendritic cells are contacted with the apoptotic cells in vitro.

18. The method according to claim 1 further comprising a maturation step following internalization of said apoptotic cells by said dendritic cells wherein said dendritic cells are exposed to a maturation factor for a sufficient time to induce maturation of said dendritic cells.

19. The method according to claim 18 wherein the maturation step comprises contacting the immature dendritic cells with at least one maturation factor selected from group consisting of monocyte conditioned medium,  $\text{TNF}\alpha$ ,  $\text{IL-1}\beta$ ,  $\text{IL-6}$ ,  $\text{PGE}_2$ ,  $\text{IFN}\alpha$ , CD40 ligand, and necrotic cells.

20. The method according to claim 19 wherein the maturation factor is selected from the group consisting of monocyte conditioned medium;  $\text{IFN}\alpha$  and at least one other factor selected from the group consisting of  $\text{IL-1}\beta$ ,  $\text{IL-6}$  and  $\text{TNF}\alpha$ ; and necrotic cells.

21. The method according to claim 20 wherein the maturation factor is necrotic cells.

22. A method of generating antigen-specific cytotoxic T lymphocytes comprising:

providing a population of apoptotic cells expressing said antigen;

- 5                   contacting dendritic cells with said apoptotic cells for a time sufficient to allow said antigen to be internalized and processed by said dendritic cells; and

- 10                   contacting T lymphocyte precursors with said dendritic cells for a sufficient time to induce the T lymphocyte precursors to become activated antigen-specific cytotoxic T lymphocytes.

- 15                   23. The method according to claim 22 wherein said dendritic cells are exposed to a preparation of apoptotic cell fragments containing antigen.

- 20                   24. The method according to claim 22 further comprising administering said antigen-specific cytotoxic T lymphocytes to an individual afflicted with a disease.

- 25                   25. The method according to claim 22 further comprising administering said apoptotic-cell primed dendritic cells to an individual afflicted with a disease for the purpose of activating antigen-specific T lymphocytes, including helper and cytotoxic T cells, in vivo.

- 30                   26. An antigen presenting dendritic cell prepared according to the method of claim 1.

27. Cytotoxic T lymphocytes prepared according to the method in claim 22.

28. A method of assessing cytotoxic T lymphocyte activity comprising:

- a) providing antigen presenting dendritic cells prepared according to the method of claim 1;
- b) exposing the antigen presenting dendritic cells to a population of T lymphocytes to be assayed for their ability to exhibit killer cell activity; and
- c) assaying the cytotoxic activity of the T lymphocytes exposed to said antigen presenting dendritic cells.

29. The method according to claim 28 wherein the antigen presented by the dendritic cells is a tumor antigen and the T lymphocytes are assayed for their cytotoxic activity against tumor cells.

30. The method according to claim 28 wherein the antigen presented by the dendritic cell is a viral antigen and the T lymphocytes are assayed for their activity against viral infected cells.

31. A method of delivering antigen to dendritic cells comprising:

contacting dendritic cells with a material selected from the group consisting of a reconstituted apoptotic cell system, apoptotic cell fragments, and liposomes comprising at least one antigen and a material which enhances internalization and translocation of antigen to an antigen processing compartment of said dendritic cells.

32. The method according to claim 31 wherein the material  
° for enhancing internalization is a ligand for the  $\alpha_v\beta_5$   
integrin receptor.

33. The method according to claim 32 wherein the ligand  
5 for the  $\alpha_v\beta_5$  integrin receptor is lactadherin.

34. The method according to claim 32 wherein the material  
for enhancing internalization is thrombospondin.

35. An in vitro culture comprising immature dendritic  
10 cells in contact with an antigen donor selected from the  
group consisting of apoptotic cells, a reconstituted  
apoptotic cell system, apoptotic cell fragments, and  
liposomes comprising at least one antigen and a material  
15 which enhances internalization and translocation of  
antigen to an antigen processing compartment of said  
dendritic cells.

36. The in vitro culture according to claim 35 wherein  
20 the dendritic cells are in contact with apoptotic cells.

37. An in vitro culture of immature dendritic cells  
wherein said dendritic cells further comprise antigen  
obtained from apoptotic cells.

25 38. An in vitro culture of mature dendritic cells  
prepared according to the method of any one of claims 19-  
22.

30 39. A pharmaceutical composition comprising dendritic  
cells which present antigen internalized from an apoptotic  
donor cell and wherein said pharmaceutical compositions

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further comprises a pharmaceutially acceptable carrier.

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40. The pharmaceutical composition according to claim 39 wherein the dendritic cells are immature.

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41. The pharmaceutical composition according to claim 39 wherein the dendritic cells are mature.

42. The pharmaceutical composition according to claim 41 wherein the internalized antigen is a tumor antigen.

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43. A method of immunizing an individual against an antigen comprising administering to an individual an amount of dendritic cells sufficient to activate T cells, wherein said dendritic cells have been contacted with antigen present on a donor selected from the group consisting of apoptotic cells, a reconstituted apoptotic cell system, apoptotic cell fragments, and liposomes comprising a material which enhances internalization and translocation of antigen to an antigen processing compartment of said dendritic cells.

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44. The method according to claim 43 wherein the dendritic cells administered to an individual are immature.

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45. The method according to claim 43 wherein the dendritic cells are contacted with a maturation factor and caused to mature prior to their administration to the individual.

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46. A method of activating CD4+ T cells comprising contacting a population of T lymphocytes with dendritic

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° cells which have been contacted with antigen present on necrotic cells and wherein the dendritic cells express antigen on MHC class II receptors.

47. The method according to claim 46 wherein the tumor is a tumor antigen.

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48. A method of activating CD4+ and CD8+ T cells against tumor cells comprising a tumor antigen, said method comprising contacting immature dendritic cells with apoptotic and necrotic cells which express said tumor antigen for a period of time sufficient for said dendritic cells to process said tumor antigen and present the tumor antigen on class I and II MHC receptors and for said dendritic cells to mature, and further contacting said antigen presenting dendritic cells with a population of T lymphocytes to cause activation of CD4+ and CD8+ T lymphocytes.

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